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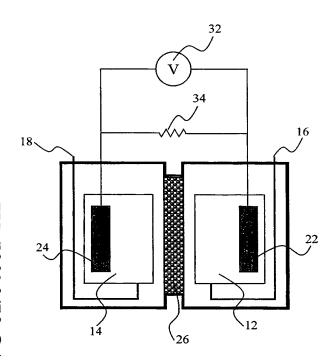
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(54) Title: A BIOFUEL CELL USING WASTEWATER AND ACTIVE SLUDGE FOR WASTEWATER TREATMENT



(57) Abstract: The present invention provides a biofuel cell using wastewater as a fuel. Electrochemically active microorganisms present in wastewater and active sludge sued in the present invention oxidize organic substances present in wastewater. Electrons generated from the oxidation are discharged outside of the microorganism cell and transferred directly to the electrode, thereby allowing electric current to be generated while allowing wastewater to be purified. The biofuel cell using the electrochemically active bacteria according to the present invention allows an electric energy of up to 0.22 mA to be generated, and also enables COD of the wastewater used as a fuel to be decreased from 1900 ppm to 55 ppm. Moreover, an efficiency of the biofuel cell is varied depending on the king and concentration of wastewater.

WO 01/04061 AJ

A BIOFUEL CELL USING WASTEWATER AND ACTIVE SLUDGE FOR WASTEWATER TREATMENT

Technical Field

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The present invention relates to a biofuel cell using wastewater as a fuel. More particularly, the present invention relates to a biofuel cell using organic substances contained in wastewater as a fuel, which biofuel cell can treat organism-containing wastewater while producing electricity. The biofuel cell according to the present invention allows reducing power generated from the catabolism of organic substances contained in wastewater by a microorganism to be converted directly into electrical energy.

A biofuel cell is a device in which an organism or its part is used and by

which reducing power generated from the energy metabolism of the organism can be

Background Art

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converted into electrical energy. In the case of a microbial fuel cell, in order to convert reducing power generated from the oxidation of a substrate by a microorganism serving as a catalyst into electrical energy, electrons generated from the energy metabolism of the microorganism should be transferred from the microorganism to an electrode. However, most of organisms including microorganisms are surrounded by a lipid membrane, a non-conductive material, at their cells. For this reason, direct electron exchange between the microorganism and the electrode cannot be effected. Therefore, when a microorganism biomass is

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used as the catalyst, a suitable electron transfer mediator should be used to facilitate electron exchange between the microorganism and the electrode. As the electron transfer mediator, an electron carrier has been used that shows a strong lipophilic property in both the oxidized form and the reduced form, and is thus capable of passing through the membrane.

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In particular, Roller et al. have proposed the use of *Proteus vulgaris*, *Escherichia coli*, *Atcaligenes eutrophus*, *Azotobacter chroococum*, or *Bacillus subtilis*, etc. as a catalyst, and thionine, methylene blue, brilliant cresyl blue, or benzyl viologen, etc. as an electron transfer mediator, in the biofuel cell (see. Roller

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et al., 1984, Journal of Chemical Technology and Biotechnology 34B: 3-12). According to Roller et al., an efficiency of the biofuel cell is significantly varied depending on the kind of the bacteria and the kind of the electron transfer mediator when being compared in view of the oxygen consumption amount.

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Moreover. Bennetto et al. have disclosed a fuel cell using sugar as a fuel, a bacterium of a Proteus genus as a catalyst, and thionine as an electron transfer mediator. The disclosed fuel cell was reported to generate up to 44 coulombs (C) of electric current (see, Bennetto et al., 1985, Biotechnology Letters, 7:699-704). Further, Robin et al. have disclosed a biofuel cell using Proteus vulgaris as a biocatalyst, 2-hydroxy-1,4-naphtoquinone (HNQ) as an electron transfer mediator, The biofuel cell according to Robin et al. has an and glucose as a fuel. electromotive power of 0.5 milliamperes (mA) and 0.7 volts (V) (see. Robin et al., 1993, Applied Biochemistry and Biotechnology 39/40:27-40). In addition, according to Habermann and Pommer, there was reported a biofuel cell that utilizes cobalt oxide or molybdenum/vanadium alloy, etc. as an electrode, and hydrogen sulfide, as a fuel, produced by sulfate-reducing bacteria contained in wastewater, and that produces 150 mA/cm² of electric current (see, Habermann and Pommer, 1991, Applied Microbiology and Biotechnology 33:128-133).

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Recently, there was separated anaerobic bacteria employing ferric ion, tetravalent manganese, hexavalent uranium, or hexavalent molybdenum, etc., as an electron receptor. Substances, that can be used as a substrate for such metal salt-reducing bacteria, include aliphatic compounds, such as lactic acid, pyruvic acid, acetic acid, propionic acid, valeric acid, and alcohol, etc., and aromatic compounds, such as toluene, phenol, cresol, benzoic acid, benzyl alchol, and benzaldehyde, etc. (see, Lovley and Klug, 1990, Applied and Environmental Microbiology 556: 1858-1864). Anaerobic bacteria are classified into fermentative bacteria and respiratory bacteria depending on their energy metabolism property. Fermentative bacteria decompose sugar and protein, etc. into organic acid, whereas respiratory bacteria completely oxidize fermentative products by the reduction of a suitable electron receptor. Electron receptors that can be used in the oxidation of organic substances by anaerobic respiratory bacteria include ferric oxide [Fe(III)], nitrate, manganese dioxide, sulfate, carbonate and the like. The reduction of ferric oxide among these

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electron receptors is known to generate the largest energy by a reducing power generated from the oxidation of a given electron donor, with the energy level being low in order of nitrate, sulfate and carbonate(see, Byoung-Hong, Kim, Microorganism Physiology, Academy Press Co., Ltd., Seoul, Korea, 1995).

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It is known that, where the iron-reducing bacteria are cultured in an anaerobic condition because of very low solubility of a ferric compound as an electron receptor in water, about 65% of their cytochromes are arranged on their outer cell membrane. By such cytochrome arrangement, reducing power generated by the oxidation of organic substances within their cells is transferred outside of the cell to reduce ferric ion outside of their cells (see, Mayers and Mayers, Journal of Bacteriology 174: 3429-3478, 1992). Moreover, it was reported that *Shewanella putrefaciens IR-1*, an iron-reducing bacterium, can generate electric current without an electron transfer mediator, by being supplied with lactic acid or hydrogen, as an electron donor (see, Park et al., 1996, Abstract, I&EC Special Symp., Sept., 16-19).

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Meanwhile, since wastewater introduced into a waste water disposal plant can contain iron at a high concentration and also ferric hydroxide is used as a phosphorus-removing agent, there will be present iron at a relatively high concentration in the wastewater disposal plant (see, Ledecke et al, 1989, Water Science and Technology 21: 325-337,). Thus, the ferric-reducing bacteria were reported to be present in most of active sludge in the wastewater disposal facility (see, Nielson et al, 1996, Water Science and Technology 34: 129-136). Also, it was reported that, in an anaerobic store condition of the active sludge, the reduction of ferric ion by microorganisms contained in the active sludge has occurred and the iron-reducing bacteria were present at a significant amount (see, Rasmussen et al., 1994, Water Research 28: 417-425).

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Based on the facts described above, where a variety of microorganisms present in active sludge or wastewater, etc. are anaerobically cultured in an anodic compartment, there will finally survive only microorganisms that are capable of employing, as an electron, an electron having a given electric potential other than the components of the culture. As a result, using such a method, electrochemically active bacteria among a variety of microorganisms present in wastewater or active sludge can be selectively densely cultured, and the respective electrochemically

PCT/KR00/00228 WO 01/04061

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active microorganisms can be isolated which are inherently present in various wastewaters.

Disclosure of the Invention

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It is therefore an object of the present invention to provide a biofuel cell that is capable of purifying wastewater while producing electricity by carrying out an efficient electrode reaction using a variety of wastewaters and sludges without using an electrode transfer mediator.

It is other object of the present invention to provide a method of treating wastewater while generating electric current by using an electrochemically active microorganism contained in wastewater and active sludge.

According to the present invention, the above objects can be accomplished by a biofuel cell comprising cathodic and anodic compartments defined in the interior of the biofuel cell and contained with conductive medium, respectively; an anode arranged in the anodic compartment; a cathode arranged in the cathodic compartment; and an ion exchange membrane interposed between the cathodic and anodic compartments and serving to divide the anodic compartment from the cathodic compartment, wherein the anodic compartment contains wastewater and active sludge.

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As described above, among microorganisms present in wastewater and active sludge contained in a biofuel cell according to the present invention, electrochemically active species grow using an electrode of a certain electric potential as an electron receptor, thereby being densely cultured. Thus, the biofuel according to the present invention is operated using the densely cultured microorganisms, as a catalyst, and organic substances present in wastewater, as a fuel.

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Brief Description of the Drawings

The above and other objects and aspects of the invention will be apparent 30 from the following description of embodiments with reference to the accompanying drawings, in which:

Fig. 1 is a schematical view showing a biofuel cell of the present invention

comprising a cathode, an anode, and a cation exchange membrane serving to divide the electrodes from each other, in which graphite felts are used as the respective electrodes.

Fig. 2 is a graph showing a reduction in electric current, electricity quantity (coulomb), and COD, which results from the use of a starch wastewater and an aerobic sludge in a biofuel cell of the present invention,

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Fig. 3 is a graph showing a reduction in electric current, electricity quantity (coulomb), and COD, which results from the use of a starch wastewater and an anaerobic sludge in a biofuel cell of the present invention,

Fig. 4 is a graph showing a reduction in electric current, electricity quantity (coulomb), and COD, which results from the use of a livestock wastewater and an anaerobic sludge in a biofuel cell of the present invention.

Fig. 5 is a graph showing a reduction in electric current, electricity quantity (coulomb), and COD, which results from the use of a wastewater from septic tank and an anaerobic sludge in a biofuel cell of the present invention,

Fig. 6a is a photograph taken with a scanning electron microscope for the surface of an electrode which is in a state before being used in a biofuel of the present invention, and

Fig. 6b is a photograph taken with a scanning electron microscope for electrochemically active microorganisms attached onto the surface of an electrode which is in a state after being used in a biofuel cell.

Best Mode for Carrying Out the Invention

Fig. 1 is a schematical view showing the structure of a biofuel cell according to the present invention. As shown in Fig. 1, the biofuel cell includes a cathodic compartment 12 and an anodic compartment 14. The cathodic and anodic compartments 12 and 14 have an oxygen introducing port 16 and a nitrogen introducing port 18, respectively. Also, in the cathodic and anodic compartments, there are arranged a cathode 22 and an anode 24, respectively. For the cathode 22 and the anode 24 of the biofuel cell, there can be used a graphite felt, a kind of graphite electrode. Moreover, in order to minimize resistance of the biofuel cell itself, a cation exchange membrane 26 is interposed between the cathodic and anodic

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compartments 12 and 14. Further, in the cathodic and anodic compartments 12 and 14, conductive media for the respective electrodes 22 and 24 are included. As the conductive medium for the cathode 22, a buffer solution is used, with the preferred buffer solution being 50 mM of phosphate buffer solution adjusted to pH 7. The cathode compartment 12 is maintained at a saturated condition by being continuously introduced with air, while the anode is maintained at an anaerobic condition by being introduced with nitrogen from which oxygen was completely removed by a passage of nitrogen through a gas oven. Additionally, in Fig. 1, reference numerals 32 and 34 represent an electrometer and a resistance terminal, respectively.

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By the anaerobic condition described above, among bacteria present in wastewater and active sludge, only microorganisms capable of using an electrode as an electron receptor can finally survive. As a result, the electrochemically active bacteria can be selectively densely cultured. The densely cultured microorganism species are used as a microorganism catalyst in the biofuel cell, such that they catabolize a variety of organic substances present in wastewater. Reducing power generated from the catabolism of the organic substances is used in the reaction with the electrode, thereby allowing electric power to be generated. Additionally, as the organic substances present in wastewater are catabolized with the densely cultured microorganisms, a concentration of the organic substances in wastewater are reduced, thereby allowing a wastewater treatment effect to be achieved.

It is preferable to use a starch wastewater and an anaerobic sludge in the anodic compartment 14 of the biofuel cell according to the present invention while using a starch wastewater and an aerobic sludge in the cathodic compartment 12. On the anodic compartment 14 that is maintained at the anaerobic condition, the densely cultured, electrochemically active bacteria produce electric current while using the organic substances in wastewater as a fuel. A cation generated from the anodic compartment 14 is passed through the cation exchange membrane 26 by which the anodic compartment 14 is divided from the cathodic compartment 12. After passing through the cation exchange membrane 26, the cation is sent to the cathodic compartment 12 saturated with oxygen, and is converted into water in the cathodic compartment 12, thereby allowing electric current to be continuously

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produced. At the same time, the organic substances present in wastewater in the cathodic compartment are catabolized with the aerobic microorganisms, such that COD of wastewater can be reduced. As a result, it is possible to treat wastewater on both the cathodic and anodic compartments 12 and 14, simultaneously.

The following examples are for further illustration purposes only and in no way limit the scope of this invention.

Example 1

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In this example, microorganisms using iron as an electron receptor among microorganisms present in wastewater contained in the biofuel cell of the present invention were measured for a change in their colony number. In this measurement, a phosphate buffer solution-based medium (PBBM) was used as a The following components were added to the medium to prepare a plate medium: 1g/L of an yeast extract, 1g/L of ammonium chloride, 25 ml/L of Macromineral (II) (including, per 1L, 6 g of KH₂PO₄, 12 g of NaCl, 2.4 g of MgSO₄ ·7H₂O, and 1.6g of CaCl₂·2H₂O), 2 ml/L of microelements (including 12.8 g of nitroacetic acid, 0.1 g of FeSO₄·7H₂O, 0.1 g of MnCl₂· 4H₂O, 0.17 g of CoCl₂·6H₄O, 0.1 g of CaCl₂·2H₂O, 0.1 g of ZnCl₂, 0.02g of CuCl₂·H₂O, 0.01 g of H₃BO₃, 0.01g of molybdate. 1.0 g of NaCl, 0.017 g of Na₂·SeO₃, and 0.026 g of NiSO₄·6H₂O), 0.1 ml/L of a vitamin solution (including 0.002 g of biotin, 0.002 g of folacin, 0.010 g of B6(pyridoxin)HCl, 0.005 g of B1(thiamin)HCl, 0.005 g of B2(riboflavin), 0.005 g of nicotinic acid(niacin), 0.005 g of panthothenic acid, 0.0001g of B12 (cyanocobalamine) crystal, 0.005 g of PABA, and 0.005 g of lipoic acid (thioctic acid)), 1ml/L of resazurin (0.2%), and 1.8% of agar.

As an electron donor, 20 mM of acetic acid, 30 mM of lactic acid, and 20 mM of glucose were used, while 20 mM of ferric pyrophosphate, a water soluble iron, was used as an electron receptor. In the first time of measurement, the respective samples of the aerobic sludge and the anaerobic sludge of the biofuel cell at the early stage of reaction were diluted with a physiological saline solution (0.8% brine) and then measured for Colony Forming Unit per ml of solution. In the second and third times, measurements were carried out using the same medium and method as in the first time, at one month and two months after the reaction, respectively. Results are shown in Table 1 below.

Table 1: Change in Colony Number in Biofuel Cell

Sample	Electron	Electron	First time	Second	Third
	donor(mM)	receptor(mM)		time	time
Aerobic	Acetic acid(20))	FP(20)	2.8×10^7	$0.9x10^4$	5.1×10^3
słudge	Glucose(20)	FP(20)	8.0×10^7	1.3×10^5	$4.2x10^4$
	Lactic acid(30)	FP(30)	6.4×10^7	1.1x10 ⁵	4.1x10 ⁴
Anaerobic	Acetic acid(20)	FP(20)	3.6x10 ⁵	5.4×10^5	1.5×10^{5}
sludge	Glucose(20)	FP(20)	2.1×10^5	8.4×10^5	$1.4x10^{5}$
	Lactic acid(30)	FP(20)	1.7x10 ⁵	1.5x10 ⁵	2.3×10^5

FP: Ferric Pyrophosphate

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As evident from Table 1 above, in the case of the aerobic sludge sample, it is believed that, as the anodic compartment of the biofuel cell is maintained in an anaerobic condition, strains other than facultative anaerobic strains are continued to reduce while being screened, such that only electrochemically active microorganisms are densely cultured. In the case of the anaerobic sludge sample, the anaerobic bacteria were increased at the second time, and then decreased at the third time, such that only electrochemically active microorganisms were densely cultured.

Example 2

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This example is to examine characteristics of a biofuel cell using a starch wastewater (collected from Samyang Genex, Co., Inchon, Korea) and an aerobic sludge (collected from Samyang Genex, Co., Inchon, Korea). For this purpose, 350 mg of a graphite felt was used for the respective electrodes of cathode and anode. As a conductive medium for the cathode, 50 mM of phosphate buffer solution was used, and the cathodic compartment and the anodic compartment were connected through a cation exchange membrane. The conductive medium for the cathodic compartment was continuously introduced with air such that it was maintained in a condition where it was saturated with oxygen. The anodic compartment was introduced with nitrogen from which oxygen has been completely

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removed by a passage of nitrogen through a gas-purifying oven. Thus, the anodic compartment was removed in dissolved oxygen such that it was maintained in an anaerobic environment. All buffer solutions used in the test were adjusted to pH 7.0. Resistance of the fuel cell was set to infinity at the early stage of the reaction. When electric pressure reached a maximum, electric current produced at a resistance of 1 k Ω was measured. A biofuel cell was used in which the aerobic sludge and the starch wastewater were mixed in the volume ratio of 1:4. The volume of the aerobic sludge and the starch wastewater contained in the biofuel cell was 25 ml in total. As electric current generated by the organic substances present in the starch wastewater was decreased. 5 ml of wastewater was replaced with fresh wastewater. The generated electric pressure was measured at an interval of 120 seconds with Potential Start Meter (2000 multimeter, keithley Instrument, Inc., USA). The measured electric pressure was divided by resistance (1k Ω) to be converted into electric current. Chemical oxygen demand (COD) of wastewater was analyzed using a standard method (see, Standard Method for the Examination of Water and Wastewater, Closed Reflux Method, 19th edition, 1995). As can be seen in Fig. 2, electric current was generated up to 0.21 mA, electricity quantity (coulomb) was increased up to 26.5 C, and COD was reduced from 1100 ppm to 58 ppm. From this experiment, it was confirmed that reducing power generated from the oxidation of a substrate in wastewater was consumed directly by an electrode to generate electric current, and also to purify the starch wastewater.

Example 3

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In this example, a biofuel cell using starch wastewater and anaerobic sludge (collected from Samyang Genex, Co., Ltd., Inchon, Korea) was tested for a electric current productivity and a wastewater treatment ability. In this test, the condition and analysis method for the biofuel cell was the same as described in Example 1.

A biofuel cell was used in which an anaerobic sludge and a starch wastewater were mixed in the volume ratio of 1:4. The volume of the anaerobic sludge and the starch wastewater contained in the biofuel cell was 25 ml in total. As can be seen Fig. 3, electric current was generated up to 0.22 mA, quantity of electricity was increased up to 26.7 Coulomb, and COD was reduced from 1940

ppm to 55 ppm. From this experiment, it was therefore confirmed that reducing power generated from the oxidation of a substrate present in starch wastewater was consumed directly by an electrode to generate electric current, and also to purify the starch waste water.

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Meanwhile, in order to examine a cultured degree of microorganisms incubated on the electrode which was used in the biofuel cell of the present invention, the electrode was photographed at its surface with an electron microscope (S-4100, FE-SEM, Hitachi, Japan) before being used in the biofuel cell. Also, after using the electrode in the biofuel cell, the electrochemically active microorganisms attached onto the electrode surface were photographed with the electron microscope. The photographed results are shown in Fig. 6a for the electrode surface and 6b for the electrochemically active microorganisms. As can be seen in Figs. 6a and 6b, it could be confirmed that the electrochemically active microorganisms were attached onto the surface of the electrode.

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Example 4

In this example, a biofuel cell was tested for an electric productivity and a wastewater treatment ability according to the same method as described in Example 2, except that a livestock wastewater (collected from Ansan Livestock, Ansan, Korea) was used instead of the starch wastewater. Also, the condition and the analysis method for the biofuel cell were the same as described in Example 1. As can seen in Fig. 4, electric current was generated up to 0.21 mA, quantity of electricity was increased up to 12 Coulombs, and COD was reduced from 1030 ppm to 350 ppm. From this experiment, it was therefore confirmed that reducing power generated from the oxidation of a substrate present in the livestock wastewater was consumed directly by an electrode to generate electric current, and also to purify the livestock wastewater.

Example 5

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In this example, a biofuel cell using a wastewater from a septic tank (collected from Apt. in Korea Institute of Science and Technology, Seoul, Korea) was tested for an electric productivity and a wastewater treatment ability. The

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operating condition and the analysis method for the biofuel cell were equal to those in Example 1. As can be seen in Fig. 5, electric current was generated up to 0.05 mA, quantity of electricity was increased up to 2.3 Coulombs, and COD was reduced from 680 ppm to 250 ppm. From this experiment, it was therefore confirmed that reducing power generated from the oxidation of a substrate in the wastewater from a septic tank was transferred directly to the electrode to generate electric current, and also to purify the wastewater from a septic tank.

Industrial Applicability

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As apparent from the above description, the present invention provides the biofuel cell utilizing wastewater and sludge. In this biofuel cell, a portion of reducing power generated when the electrochemically active microorganisms contained in the sludge are subjected to the energy metabolism with the substrate present in wastewater, is utilized for the production of a biomass. At the same time, the remaining portion of the reducing power is utilized to produce electric current while purifying wastewater. As a result, where the biofuel cell utilizes a variety of wastewaters as a fuel, it then can achieve the electrical energy production and the wastewater treatment effect, simultaneously.

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Although the preferred embodiments of the invention have been disclosed for illustrative purposes, those skilled in the art will appreciate that various modifications, additions and substitutions are possible, without departing from the scope and spirit of the invention as disclosed in the accompanying claims.

WO 01/04061

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Claims

1. A biofuel cell comprising cathodic and anodic compartments defined in the interior of the biofuel cell and contained with conductive medium, respectively; an anode arranged in the anodic compartment; a cathode arranged in the cathodic compartment; and an ion exchange membrane interposed between the cathodic and anodic compartments and serving to divide the anodic compartment from the cathodic compartment, wherein the anodic compartment contains wastewater and active sludge and is maintained in an anaerobic condition during an operation of the biofuel cell.

- 2. The biofuel cell of Claim 1, in which the active sludge and the wastewater are selected from the group consisting of a starch wastewater, a livestock wastewater, a wastewater from a septic tank, and a combination thereof.
- 3. The biofuel cell of Claim 1. in which the cathodic compartment contains the sludge and the wastewater.
- 4. A method of treating wastewater while producing electric power using the biofuel cell of Claim 1. comprising of:

introducing the wastewater and the active sludge into the anodic compartment of the biofuel cell;

introducing nitrogen into the anodic compartment to remove dissolved oxygen from the anodic compartment, such that the anodic compartment is maintained in an anaerobic condition,

continuously introducing air into the cathodic compartment, such that the cathodic compartment is maintained in a condition where it is saturated with oxygen, and

densely culturing electrochemically active microorganisms present in the wastewater and the active sludge,

whereby the cultured active bacteria are used as a microorganism catalyst, and organic substances present in the wastewater are used as a fuel.

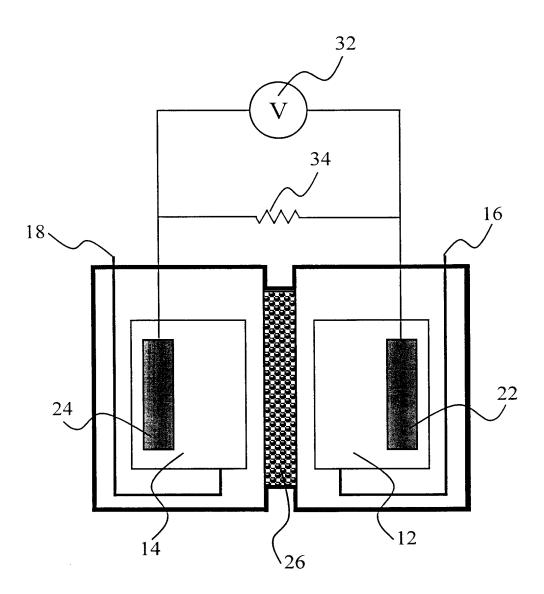
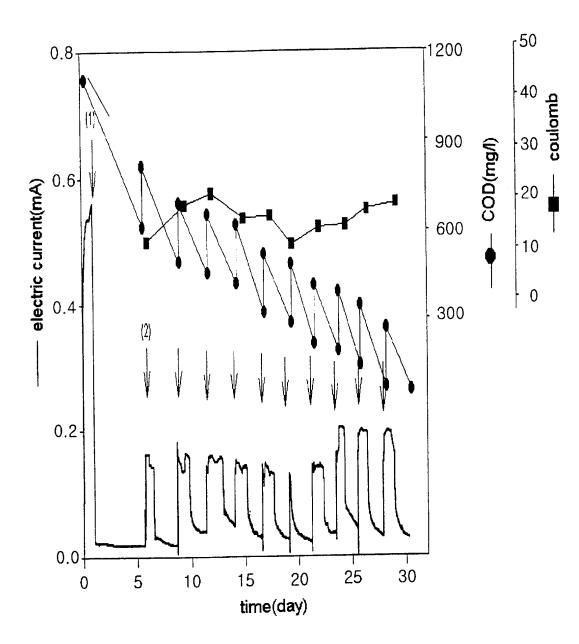


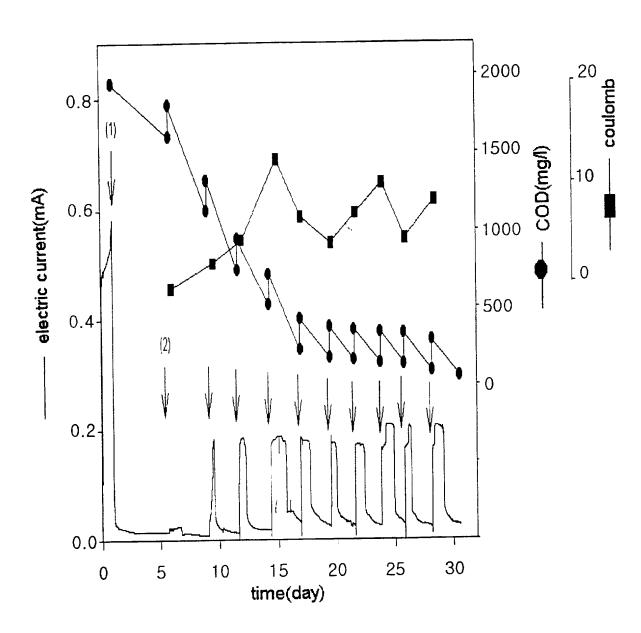
Fig. 1

WO 01/04061



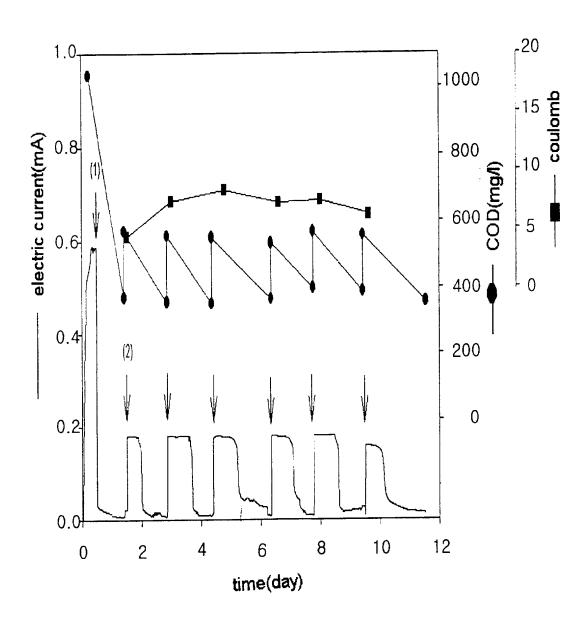
(1) electric discharge (2) starch wastewater supply

Fig. 2.



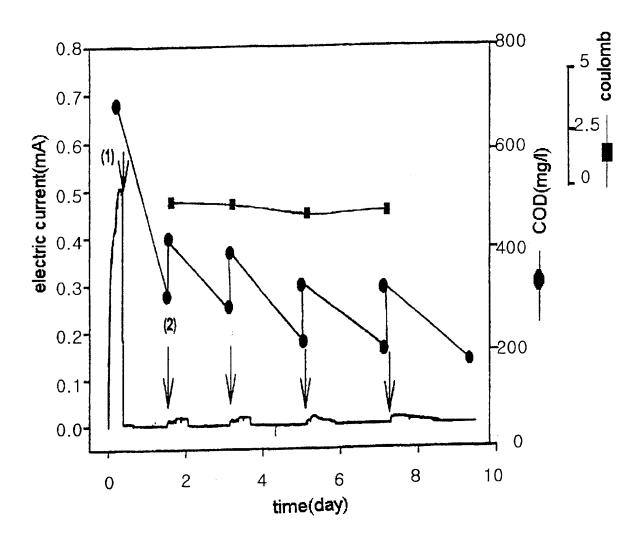
(1) electric discharge (2) starch wastewater supply

Fig. 3.



(1) electric discharge (2) livestock wastewater supply

Fig. 4.



(1) electric discharge (2) septic tank-wastewater supply

Fig. 5.

PCT/KR00/00228

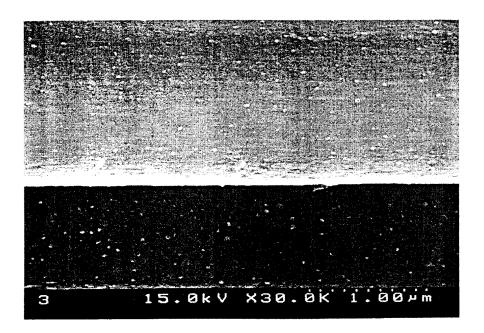


Fig. 6a.

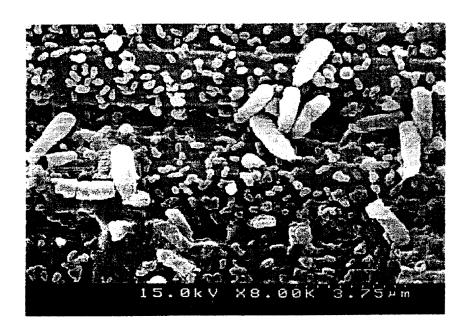


Fig. 6b.

INTERNATIONAL SEARCH REPORT

international application No. PCT/KR00/00228

Α.	CLASSIFICATION	OF	SUBJECT	MATTER
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IPC7 C02F 3/00, H01M 8/16

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimun documentation searched (classification system followed by classification symbols)

IPC7 C02F 3/00, H01M 8/16, C02F 1/46

Documentation searched other than minimun documentation to the extent that such documents are included in the fileds searched

Korean Patents and applications for inventions since 1975

Korean Utility models and applications for Utility models since 1975

Electronic data base consulted during the intertnational search (name of data base and, where practicable, search trerms used) IPN, NPS, PAJ, CA

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	US 5702835 A (Ross Carson Larue) 30 Dec 1997 see the whole document,	1-4
A	JP 4-2058 A (Kubota Corp.) 7 Jan 1992 see the whole document.	1, 3
A	KR 98-16777 A (Korea Institute of Science and Technology) 5 Jun 1998 see Fig. 1	1

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